L3 ANSWER 11 OF 11 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1997:140289 CAPLUS
DOCUMENT NUMBER: 126:139879
TITLE: Gene therapy of solid tumors with interferons alone or with other immuno-effector proteins
INVENTOR(S): Pestka, Sidney; Sarkar, Srijata; Flores, Idhaliz; Ron, Yacov
PATENT ASSIGNEE(S): University of Medicine & Dentistry of New Jersey, USA
SOURCE: PCT Int. Appl., 77 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9700085	A1	19970103	WO 1996-US10502	19960618
W: AL, AM, AU, BB, BG, BR, BY, CA, CN, CZ, EE, FI, GE, HU, IL, IS, JP, KG, KP, KR, LK, LR, LT, LV, MD, MG, MK, MN, MX, NO, NZ, PL, RO, SG, SI, SK, TR, TT, UA, UZ, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9662828	A1	19970115	AU 1996-62828	19960618
EP 835130	A1	19980415	EP 1996-921666	19960618
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				

PRIORITY APPLN. INFO.: US 1995-302P P 19950619
WO 1996-US10502 W 19960618

AB **Vectors** and compns. are provided for gene therapy of solid tumors, particularly malignant tumors. A vaccine for treatment of solid tumors comprises tumor cells transfected to express interferon-.alpha. in a pharmaceutically acceptable excipient. Preferably, the tumor cells are also transfected to express an immunomodulatory mol., such as interferon-.gamma., interferon-.beta., interferon-.omega., interferon-.tau., tumor necrosis factor-.alpha., tumor necrosis factor-.beta., interleukin-2, interleukin-7, interleukin-12, interleukin-15, **B7**-1 T-cell **costimulatory mol**., **B7**-2 T-cell **costimulatory mol**., immune cell adhesion mol. (**ICAM**)-1 T-cell **costimulatory mol**., granulocyte colony-stimulatory factor, granulocyte-macrophage colony-stimulatory factor, and combinations thereof, with the proviso that the immunomodulatory mol. is not interferon-.alpha.. A sol. immunomodulatory mol. can be included in the vaccine. Solid tumors are treated by introducing into the patient a therapeutically effective no. of solid tumor cells which are transfected to express interferon-.alpha. and preferably an addnl. immunomodulatory mol. Alternatively, a therapeutically effective no. of addnl. tumor cells transfected to express an immunomodulatory mol. can be introduced into the subject.

```
=> vector (l) costimulatory (w) molecule
         121476 VECTOR
          68802 VECTORS
         162532 VECTOR
                  (VECTOR OR VECTORS)
           3708 COSTIMULATORY
          41785 MOLECULE
         119910 MOLECULES
         156675 MOLECULE
                  (MOLECULE OR MOLECULES)
        1976070 MOL
         529664 MOLS
        2264909 MOL
                  (MOL OR MOLS)
        2297289 MOLECULE
                  (MOLECULE OR MOL)
L1          114 VECTOR (L) COSTIMULATORY (W) MOLECULE

=> B7 (s) ICAM
           9744 B7
           9648 ICAM
             87 ICAMS
           9662 ICAM
                  (ICAM OR ICAMS)
L2          275 B7 (S) ICAM

=> L1 and L2
L3           11 L1 AND L2

=> LFA (s) B7
           4355 LFA
             19 LFAS
           4365 LFA
                  (LFA OR LFAS)
           9744 B7
L4          132 LFA (S) B7

=> L1 and L4
L5            9 L1 AND L4

=> L2 and L4
L6           98 L2 AND L4

=> L1 and L6
L7            8 L1 AND L6

=> CD40 (s) B7
           4775 CD40
           9744 B7
L8          324 CD40 (S) B7

=> L1 and L8
L9            3 L1 AND L8

=> B7 (s) CD59
           9744 B7
            894 CD59
L10           3 B7 (S) CD59

=> B7 (l) CD59
```

```
          9744 B7
           894 CD59
L11          4 B7 (L) CD59

=> L1 and L11
L12          0 L1 AND L11

=> B7 (l) CD70
          9744 B7
           128 CD70
L13          7 B7 (L) CD70

=> L1 and L13
L14          0 L1 AND L13

=> B7 (l) OX-40L
          9744 B7
         12849 OX
             2 OXES
           468 OXEN
         13236 OX
                 (OX OR OXES OR OXEN)
           137 40L
             9 OX-40L
                 (OX(W)40L)
L15          2 B7 (L) OX-40L

=> L1 and L15
L16          0 L1 AND L15

=> B7 (l) VCAM
          9744 B7
          3862 VCAM
             2 VCAMS
          3862 VCAM
                 (VCAM OR VCAMS)
L17         48 B7 (L) VCAM

=> L1 and L17
L18          0 L1 AND L17

=> B7 (l) ICAM (l) LFA
          9744 B7
          9648 ICAM
            87 ICAMS
          9662 ICAM
                 (ICAM OR ICAMS)
          4355 LFA
            19 LFAS
          4365 LFA
                 (LFA OR LFAS)
L19        135 B7 (L) ICAM (L) LFA

=> L1 and L19
L20          9 L1 AND L19
```

L20 ANSWER 1 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:278014 CAPLUS
TITLE: Antitumor Immunity After Vaccination With B Lymphoma Cells Overexpressing a Triad of Costimulatory Molecules
AUTHOR(S): Briones, Javier; Timmerman, John M.; Panicalli, Dennis L.; Levy, Ronald
CORPORATE SOURCE: CA; D. L. Panicalli, Stanford, Division of Oncology, R. Levy, J. M. Timmerman, J. Briones, Stanford University School of Medicine, Therion Biologics Corporation, Cambridge, MA, USA
SOURCE: Journal of the National Cancer Institute (2003), 95(7), 548-555
CODEN: JNCIEQ; ISSN: 0027-8874
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Background: The **costimulatory mols.** **B7**-1, intercellular adhesion mol.-1 (**ICAM**-1), and leukocyte function-assocd. antigen-3 (**LFA**-3) play pivotal roles in the activation of T cells. We investigated whether in vivo vaccination with lymphoma cells infected with a recombinant, nonreplicating fowlpox (FP) virus encoding this triad of **costimulatory mols.** (TRICOM) could stimulate lymphoma-specific immunity. Methods: TRICOM-infected A20 B lymphoma cells were analyzed for expression of **B7**-1, **ICAM**-1, and **LFA**-3. Mice (10 per group) were vaccinated with irradiated A20 cells infected with either the TRICOM **vector** or the wild-type FP virus (WT-FP), challenged with live A20 tumor cells, and followed for survival. Mice with established A20 tumors were also treated with irradiated TRICOM-infected A20 cells. Survival curves were compared with the log-rank statistic. The mechanism of the antitumor effect was studied by in vivo depletion of CD4+ and CD8+ T cells and in vitro cytotoxicity assays. All statistical tests were two-sided. Results: A20 tumor cells infected with TRICOM expressed high levels of **B7**-1, **ICAM**-1, and **LFA**-3. Mice vaccinated with irradiated TRICOM-infected A20 cells had prolonged survival relative to mice vaccinated with WT-FP-infected cells (80% vs. 20% survival at 110 days; P<.001). In mice with established tumors, tumor growth was slower in those treated with TRICOM-infected tumor cells than in those treated with WT-FP-infected cells, and this treatment provided a survival advantage (P<.001). Depletion of CD4+ or CD8+ T cells reduced the antitumor immunity provided by the tumor cell-TRICOM vaccine, and lymphocytes from vaccinated mice displayed in vitro cytotoxic activity toward A20 cells. Conclusions: Increasing expression of **costimulatory mols.** on B lymphoma cells by infection with a recombinant FP virus encoding **B7**-1, **ICAM**-1, and **LFA**-3 stimulates antitumor immune responses in vivo and may provide a novel strategy for treating patients with B-cell malignancies.

L20 ANSWER 2 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:129253 CAPLUS
DOCUMENT NUMBER: 138:253285
TITLE: Selective Induction of High Avidity CTL by Altering the Balance of Signals from APC
AUTHOR(S): Oh, SangKon; Hodge, James W.; Ahlers, Jeffrey D.; Burke, Donald S.; Schlom, Jeffrey; Berzofsky, Jay A.
CORPORATE SOURCE: National Cancer Institute, and Laboratory of Tumor Immunology and Biology, Metabolism Branch, Molecular

Immunogenetics and Vaccine Research Section, National Institutes of Health, Bethesda, MD, 20892, USA
SOURCE: Journal of Immunology (2003), 170(5), 2523-2530
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English

AB High avidity CTL are most effective at clearing viruses and cancer cells. Therefore, understanding the mechanisms involved in induction of high avidity CTL is crit. for effective vaccines. However, no vaccine approach to selectively induce high avidity CTL in vivo has been discovered. In a new approach, signals from MHC class I (signal 1) and costimulatory mols. (signal 2) were adjusted by varying Ag dose and by use of recombinant poxvirus expressing a triad of costimulatory mols. (**B7**-1, **ICAM**-1, and **LFA**-3), resp. Independent of CTL avidity, a strong signal 1 resulted in an increased frequency of CD8+ CTL. However, a strong signal 2 was necessary for the induction of high avidity CD8+ CTL that killed target cells more efficiently, and signal 2 played a more crucial role in the absence of a strong signal 1. Only CTL induced with strong signal 2 killed tumor cells endogenously expressing low levels of Ag. Signal 2 contributed to the induction of high avidity CD8+ CTL in both primary and secondary responses. Thus, although signal 2 has been known to increase the quantity of CTL response, in this study the authors show that it also improves the quality of CTL response. The data also suggested that dendritic cells play an important role in induction of high avidity CD8+ CTL in vivo. This strategy to selectively induce higher avidity CTL may lead to more effective vaccines for viruses and cancer.

REFERENCE COUNT: 40 THERE ARE 40 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 3 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:900432 CAPLUS
DOCUMENT NUMBER: 137:77486
TITLE: Enhanced activation of rhesus T cells by **vectors** encoding a triad of **costimulatory molecules** (**B7**-1, **ICAM**-1, **LFA**-3)
AUTHOR(S): Shankar, Pragyna; Schlom, Jeffrey; Hodge, James W.
CORPORATE SOURCE: Research Scholar's Program, NIH, Howard Hughes Medical Institute, Bethesda, MD, 20892, USA
SOURCE: Vaccine (2001), 20(5-6), 744-755
CODEN: VACCDE; ISSN: 0264-410X
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Since the rhesus is often used as a "gatekeeper" model for the evaluation of malaria and simian immunodeficiency virus (SIV)/HIV vaccines, the identification of strategies to enhance the activation of rhesus T cells would potentially aid in the generation of more potent vaccines directed against these infectious agents. Several mols. normally found on the surface of professional human APCs are capable of providing the second signals crit. for T cell activation: **B7**-1 (CD80), **ICAM**-1 (CD54), and **LFA**-3 (CD58). With the exception of **B7**, T cell **costimulatory mols**. in the rhesus have not

been identified. We have recently designed and characterized both recombinant vaccinia and recombinant avipox **vectors** contg. the transgenes for a triad of human T cell **costimulatory mols**. (**B7**-1, **ICAM**-1, **LFA**-3; designated TRICOM). Here, we demonstrate the enhanced activation of rhesus T cells stimulated with rhesus APCs infected with TRICOM **vectors** in the presence of signal 1. Infection with TRICOM **vectors** led to significant improvement of APC capabilities in terms of redn. of the amt. of signal 1 needed to activate naive T cells, and redn. in the amt. of APCs required to activate T cells using a const. amt. of signal 1. Antibody blocking studies demonstrated that each of the three **costimulatory mol**. transgenes contributed to the enhanced proliferation of T cells. TRICOM-enhanced T cell activation was shown to correspond to increases in type 1 cytokines and a reduced level of apoptosis. TRICOM-infected autologous B cells from rhesus immunized with either an SIV vaccine or a malaria vaccine stimulated significantly greater levels of IFN-.gamma. in response to specific peptide than stimulation with uninfected autologous B cells or B cells infected with wild-type **vector**. The ability to augment immune responses using poxvirus-based vaccines contg. multiple **costimulatory mol**. transgenes can now be addressed in the rhesus macaque model.

REFERENCE COUNT: 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 4 OF 9 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:683489 CAPLUS
DOCUMENT NUMBER: 136:245917
TITLE: Technology evaluation: CEA-TRICOM, Therion Biologics Corp
AUTHOR(S): Morse, Michael A.
CORPORATE SOURCE: Department of Medicine, Duke University Medical Center, Durham, NC, 27710, USA
SOURCE: Current Opinion in Molecular Therapeutics (2001), 3(4), 407-412
CODEN: CUOTFO; ISSN: 1464-8431
PUBLISHER: PharmaPress Ltd.
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review. Therion Biologics, the NCI and Aventis Pasteur are investigating CEA-TRICOM, a recombinant, pox virus-based vaccine that incorporates a triple dose of **costimulatory mols**. as well as the carcinoembryonic antigen (CEA) tumor antigen, for the potential treatment of colorectal cancer. CEA-TRICOM is designed to stimulate and strengthen the body's immune system to kill colorectal cancer cells. CEA-TRICOM is administered in a priming and boosting protocol using two unique pox virus **vectors**, rV-CEA-TRICOM (recombinant vaccinia **vector**) and rF-CEA-TRICOM (recombinant fowlpox **vector**). The TRICOM component of both rV-CEA-TRICOM and rF-CEA-TRICOM comprises three **costimulatory mol**. transgenes (**B7**-1, **ICAM**-1 and **LFA**-3) known to elicit strong cellular immune responses necessary for complete tumor destruction. In preclin. studies conducted by the NCI and Therion, researchers have demonstrated that this combination of three **costimulatory mols**. dramatically boosts the immune response to eradicate cancer in murine models. In Feb. 2001, Therion Biologics and the NCI initiated a phase I trial of CEA-TRICOM. The phase I trial of CEA-TRICOM is designed to demonstrate proof-of-principle for

using multiple **costimulatory mols**. in conjunction with a tumor antigen to improve the strength of cellular immune responses. It is a multistage, dose-escalation study that will assess the safety and immunol. effects of CEA-TRICOM in up to 42 patients who have advanced metastatic colorectal cancer. Subjects will receive rF-CEA-TRICOM alone, rV-CEA-TRICOM followed by booster vaccinations with rF-CEA-TRICOM or rV-CEA-TRICOM followed by rF-CEA-TRICOM and GM-CSF adjuvant. The primary measure of immune response will be the level of CEA-specific T-cells stimulated by vaccination, with levels of CEA-expressing tumor cells in the blood used as a potential secondary measure of treatment effect.

REFERENCE COUNT: 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 5 OF 9 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:431014 CAPLUS
DOCUMENT NUMBER: 135:179366
TITLE: Synergy of vaccine strategies to amplify antigen-specific immune responses and antitumor effects
AUTHOR(S): Grosenbach, Douglas W.; Barrientos, Jacqueline C.; Schlom, Jeffrey; Hodge, James W.
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892-1750, USA
SOURCE: Cancer Research (2001), 61(11), 4497-4505
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Several different vaccine strategies have been evaluated and combined to amplify T-cell responses toward induction of antitumor immunity. The model tumor antigen used was carcinoembryonic antigen (CEA). While initial T-cell activation studies were conducted in conventional mice, combined vaccine strategy studies and antitumor studies were conducted in transgenic mice in which CEA is expressed in normal gastrointestinal tissue and CEA protein is found in sera. The studies reported here demonstrate: (a) A recombinant avipox (fowlpox, rF) **vector** expressing the signal 1 (CEA) and the **B7**-1 **costimulatory mol**. transgenes (designated rF-CEA/**B7**-1) is more potent in inducing CEA-specific T-cell responses than rF-CEA; one administration of recombinant fowlpox **vector** expressing CEA and three different **costimulatory mol**. transgenes (**B7**-1, **ICAM**-1, **LFA**-3, designated rF-CEA/TRICOM) was more potent in inducing CEA-specific T-cell responses than four vaccinations with rF-CEA or two vaccinations with rF-CEA/**B7**-1. Moreover, up to four vaccinations with rF-CEA/TRICOM induced greater CEA-specific T-cell responses with each vaccination. (b) A diversified prime and boost strategy using a prime with a recombinant vaccinia **vector** expressing CEA and the triad of **costimulatory mols**. (designated rV-CEA/TRICOM) and a boost with rF-CEA/TRICOM was more potent in inducing CEA-specific T-cell responses than the repeated use of rF-CEA/TRICOM alone. (c) The addn. of granulocyte macrophage colony-stimulating factor (GM-CSF) to the rF-CEA or rF-CEA/TRICOM vaccinations via the simultaneous administration of a rF-GM-CSF **vector** enhanced CEA-specific T-cell responses. These strategies (TRICOM/diversified prime and boost/GM-CSF) were combined to treat CEA-expressing carcinoma liver metastases in CEA-transgenic mice; vaccination was initiated 14 days post-tumor transplant. Antitumor effects in terms of survival and CD8+ and CD4+ responses specific for CEA

were also obsd. in this CEA-transgenic mouse model. These studies demonstrate that the use of cytokines and diversified prime and boost regimens can be combined with the use of recombinant **vectors** expressing signal 1 and multiple **costimulatory mols.** to further amplify T-cell responses toward more effective vaccine strategies.

REFERENCE COUNT: 72 THERE ARE 72 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 6 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:354386 CAPLUS
DOCUMENT NUMBER: 135:120924
TITLE: Enhanced activation of human T cells via avipox **vector**-mediated hyperexpression of a triad of **costimulatory molecules** in human dendritic cells
AUTHOR(S): Zhu, MingZhu; Terasawa, Hiroshi; Gulley, James; Panicali, Dennis; Arlen, Philip; Schlom, Jeffrey; Tsang, Kwong Y.
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892-1750, USA
SOURCE: Cancer Research (2001), 61(9), 3725-3734
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB T-cell activation usually requires at least 2 signals. The first signal is antigen-specific, and the second signal(s) involves the interaction of a T-cell **costimulatory mol.**(s) on the antigen-presenting cell (APC) with its ligand on the T cell. Dendritic cells (DCs) are the most potent APCs, attributable, in part, to their expression of several T-cell **costimulatory mols.** Human DCs generated in vitro, however, will vary in methods of generation and maturation and in terms of expression of different phenotypic markers-including **costimulatory mols.**-among different donors. The authors report here that a recombinant avipox (fowlpox, rF) **vector** has been constructed that can efficiently express the transgenes for 3 human T-cell **costimulatory mols.** (**B7**-1, **ICAM**-1, and **LFA**-3) as a result of individual early avipox promoters driving the expression of each transgene. This triad of **costimulatory mols.** (designated TRICOM) was selected because each has an individual ligand on T cells and each has been shown previously to prime a unique signaling pathway in T cells. The authors report here that rF-TRICOM can efficiently infect human DCs of different states of maturity and hyperexpress each of the 3 **costimulatory mols.** on the DC surface without affecting other DC phenotypic markers. Infection of influenza or human papilloma virus 9-mer peptide-pulsed DCs from different individuals, or at different stages of maturity with rF-TRICOM, resulted in enhanced activation of T cells from peripheral blood mononuclear cells of autologous donors after 24 h of incubation with DCs. This enhanced activation was analyzed by both titrating the peptide and differing the DC:effector cell ratios. No effect was obsd. using the control wild-type avipox **vector.** No increase in apoptosis was obsd. in T cells hyperstimulated with the TRICOM **vector,** and no decrease in interleukin-12 prodn. was seen in lipopolysaccharide-stimulated DCs infected with rF-TRICOM. Antibody-blocking expts. demonstrated that

enhanced T-cell activation by TRICOM was attributed to each of the 3 **costimulatory mols**. Peptide-pulsed, rF-TRICOM-infected DCs were also shown to be more effective than peptide-pulsed DCs in activating T cells to 9-mer peptides derived from 2 relatively weak "self" immunogens, i.e., human prostate-specific antigen and human carcinoembryonic antigen. These studies thus demonstrate for the first time that a **vector** that can simultaneously hyperexpress 3 **costimulatory mols**. can be used to efficiently infect human DCs, leading to enhanced peptide-specific T-cell activation. The use of this approach for in vitro studies and clin. applications in immunotherapy is discussed.

REFERENCE COUNT: 62 THERE ARE 62 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 7 OF 9 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:334023 CAPLUS
DOCUMENT NUMBER: 136:149737
TITLE: Enhancing the potency of peptide-pulsed antigen presenting cells by **vector**-driven hyperexpression of a triad of **costimulatory molecules**
AUTHOR(S): Hodge, J. W.; Grosenbach, D. W.; Rad, A. N.; Giuliano, M.; Sabzevari, H.; Schlom, J.
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, National Institutes of Health, Bethesda, MD, 20892-1750, USA
SOURCE: Vaccine (2001), 19(25-26), 3552-3567
CODEN: VACCDE; ISSN: 0264-410X
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Recombinant orthopox **vectors** [both replication-defective fowlpox (rF), and replication competent vaccinia (rV)] have been developed that simultaneously express 3 T-cell **costimulatory mol**. transgenes. The constituents of this triad of **costimulatory mols**. (designated TRICOM) are **B7**-1, **ICAM**-1, and **LFA**-3. The authors have previously shown that infection of murine dendritic cells (DCs) with TRICOM **vectors** increases their level of expression of the triad of **costimulatory mols**. and enhances the efficacy of DCs to activate T cells. While DCs are arguably the most potent antigen presenting cell (APC), limitations clearly exist in their use due to the level of effort and cost for their generation. The studies reported here demonstrate that a generic APC population, murine splenocytes, can be made markedly more efficient as APCs by infection with either rF-TRICOM or rV-TRICOM **vectors**. Infection of splenocytes with either TRICOM **vector** led to improvement of APC capabilities in terms of: (1) enhancement of mixed lymphocyte reactions; (2) a redn. in the amt. of signal 1 to activate naive T cells; and (3) a redn. in the amt. of APCs required to activate T cells using a const. amt. of signal 1. TRICOM-enhanced T-cell activation was shown to correspond to increases in type-1 cytokines and a reduced level of apoptosis, compared with T cells activated with uninfected or control **vector**-infected splenocytes. In vitro and in vivo expts. compared DCs with TRICOM-infected splenocytes. Infection of splenocytes with TRICOM **vectors** markedly enhanced their ability to activate T cells to levels approaching that of DCs. These studies thus

demonstrate for the first time that an abundant and accessible population of APCs obtainable without lengthy culture or the use of costly exogenous cytokines (in contrast to that of DCs) can be made more potent as APCs with the use of **vectors** that express a triad of **costimulatory mols**.

REFERENCE COUNT: 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 8 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1999:778296 CAPLUS
DOCUMENT NUMBER: 132:77584
TITLE: A triad of costimulatory molecules synergize to amplify T-cell activation
AUTHOR(S): Hodge, James W.; Sabzevari, Helen; Yafal, Alicia Gomez; Gritz, Linda; Lorenz, Matthias G. O.; Schlom, Jeffrey
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892, USA
SOURCE: Cancer Research (1999), 59(22), 5800-5807
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: AACR Subscription Office
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The activation of a T cell has been shown to require 2 signals via mols. present on professional antigen-presenting cells: signal 1, via a peptide/MHC complex; and signal 2, via a **costimulatory mol**. Here, the role of 3 **costimulatory mols**. in the activation of T cells was examd. Poxvirus (vaccinia and avipox) **vectors** were used because of their ability to efficiently express multiple genes. Murine cells provided with signal 1 and infected with either recombinant vaccinia or avipox **vectors** contg. a TRIad of **COstimulatory Mols**. (**B7**-1/**ICAM**-1/**LFA**-3, designated TRICOM) induced the activation of T cells to a far greater extent than cells infected with any 1 or 2 **costimulatory mols**. Despite this T-cell "hyperstimulation" using TRICOM **vectors**, no evidence of apoptosis above that seen using the **B7**-1 **vector** was obsd. Results using the TRICOM **vectors** were most dramatic under conditions of either low levels of first signal or low stimulator cell:T-cell ratios. Expts. using a 4-gene construct also showed that TRICOM recombinants can enhance antigen-specific T-cell responses in vivo. These studies thus demonstrate for the first time the ability of **vectors** to introduce 3 **costimulatory mols**. into cells, thereby activating both CD4+ and CD8+ T-cell populations to levels greater than those achieved with the use of only 1 or 2 **costimulatory mols**. This new threshold of T-cell activation has broad implications in vaccine design and development.

REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L20 ANSWER 9 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1999:187281 CAPLUS
DOCUMENT NUMBER: 130:350899
TITLE: Induction of anti-tumor immunity elicited by tumor cells expressing a murine LFA-3 analog via a

recombinant vaccinia virus

AUTHOR(S): Lorenz, Matthias G. O.; Kantor, Judy A.; Schlom, Jeffrey; Hodge, James W.

CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, Bethesda, MD, 20892, USA

SOURCE: Human Gene Therapy (1999), 10(4), 623-631
CODEN: HGTHE3; ISSN: 1043-0342

PUBLISHER: Mary Ann Liebert, Inc.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB T cell activation requires binding of the T cell receptor to the major histocompatibility mol.-peptide complex in the presence of adhesion and/or **costimulatory mols.** such as **B7**-1 (CD80), **B7**-2 (CD86), **ICAM**-1 (CD54), and CD2. The major ligand of CD2 is CD48, the murine analog of human leukocyte function-assocd. antigen 3 (**LFA**-3). To det. the effect of **LFA**-3 expression on the immunogenicity of tumor cells, the authors constructed a recombinant vaccinia virus contg. the murine **LFA**-3 gene (designated rV-**LFA**-3). RV-**LFA**-3 was shown to be functional in vitro in terms of expression of **LFA**-3, T cell proliferation, adhesion, and cytotoxicity. S.c. inoculation of rV-**LFA**-3-infected murine colon adenocarcinoma tumor cells (MC38) into immunocompetent syngeneic C57BL/6 mice resulted in complete lack of tumor growth. Inoculation of MC38 cells infected with equal doses of control wild-type vaccinia virus resulted in tumor growth in all animals. In addn., partial immunol. protection was demonstrated against subsequent challenge with uninfected parental tumor cells up to 56 days after vaccination with rV-**LFA**-3-infected cells. Anti-tumor memory was also demonstrated by using .gamma.-irradiated MC38 cells and cells from another carcinoma model (CT26). These studies demonstrate that expression of **LFA**-3 via a poxvirus **vector** can be used to induce anti-tumor immunity.

REFERENCE COUNT: 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 2 OF 2 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1998:811923 CAPLUS
DOCUMENT NUMBER: 130:166899
TITLE: Ox-40 ligand: a potent costimulatory molecule for sustaining primary CD4 T cell responses
AUTHOR(S): Gramaglia, Irene; Weinberg, Andrew D.; Lemon, Michael; Croft, Michael
CORPORATE SOURCE: Div. Immunochem., La Jolla Inst. Allergy Immunol., San Diego, CA, 92121, USA
SOURCE: Journal of Immunology (1998), 161(12), 6510-6517
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Ox-40 and Ox-40 ligand (**Ox-40L**) are thought to be involved in T cell-APC interactions. However, their exact role in T cell responses is undefined. Using fibroblast transfectants expressing **Ox-40L** and/or **B7**-1, and CD4 cells from TCR transgenic mice, we investigated the effect of Ox-40 signaling on primary responses to the Ag pigeon cytochrome c. Ox-40 expression on naive CD4 cells peaked 2 to 3 days after activation, and was lost by 4 to 5 days. APCs with **Ox-40L** promoted partial activation of naive T cells with some IL-2 secretion, but were unable to enhance proliferation, unlike those with **B7**-1. APCs coexpressing **Ox-40L** with **B7**-1 induced large quantities of IL-2 and promoted proliferative responses that persisted for several days. Effector cells taken 5 days after naive T cell activation reexpressed Ox-40 within 4 h and responded strongly to APCs expressing **Ox-40L**, whereas **B7**-1 had little effect. Synergy was also seen between **Ox-40L** and **B7**-1, with primarily IL-2 being elevated, although IL-4 and IL-5 were also up-regulated. The most striking action was on effector T cell proliferation, which continued at high levels for up to 4 days, with little proliferation evident at this time in the absence of Ox-40 signals. These data suggest that Ox-40/**Ox-40L** interactions act after initial activation events to prolong clonal expansion and enhance effector cytokine secretion, and may be involved in promoting long-lived primary CD4 responses.

REFERENCE COUNT: 58 THERE ARE 58 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 1 OF 7 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:216623 CAPLUS
TITLE: Involvement of CD70 and CD80 intracytoplasmic domains in the co-stimulatory signal required to provide an antitumor immune response
AUTHOR(S): Douin-Echinard, Victorine; Peron, Jean-Marie; Lauwers-Cances, Valerie; Favre, Gilles; Couderc, Bettina
CORPORATE SOURCE: Inserm U563, CPTP, Laboratoire d'Innovation Therapeutique et Oncologie Moleculaire, Institut Claudius Regaud, 20-24 rue du pont St Pierre, Toulouse, 31052, Fr.
SOURCE: International Immunology (2003), 15(3), 359-372
CODEN: INIMEN; ISSN: 0953-8178
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English

AB **CD70** and CD80 are co-stimulatory mols. which belong to the tumor necrosis factor family and the **B7** family resp. When they are co-expressed by gene-modified TS/A tumor cells, they provide an efficient protective and long-lasting T-dependent antitumor response. We first showed that when **CD70** and CD80 were delivered in the tumor environment by gene-modified fibroblasts, but were not expressed by the tumor cells themselves, no antitumor response was obsd. We next assessed whether the intracytoplasmic domains of **CD70** and CD80 contribute to enhance the co-stimulatory activity necessary to induce effective T cell-tumor cell interactions and T cell-dependent antitumor response. TS/A cells were gene-modified to express different combinations of deleted (**CD70**.DELTA. and CD80.DELTA.) or full-length **CD70** and CD80 co-stimulatory mols. In vitro, the CD80 intracytoplasmic domain was required to regulate CD80 membrane redistribution by interacting with the actin cytoskeleton. The loss of the **CD70** intracytoplasmic domain did not alter its ability to relocate on the surface membrane, but failed to co-stimulate T cell proliferation. In vivo expts. in syngeneic BALB/c mice showed that the **CD70**/CD80-TS/A and the **CD70**.DELTA./CD80-TS/A tumors were rejected via CD8 T cells, whereas **CD70**/CD80.DELTA.-TS/A and **CD70**.DELTA./CD80.DELTA.-TS/A tumors were not. The mice that rejected **CD70**.DELTA./CD80-TS/A tumors showed decreased protection against injection of parental TS/A cells when compared to mice which rejected **CD70**/CD80-TS/A tumors. These results showed that the intracytoplasmic domain of CD80 was crit. for the effector phase of CD8 T cell-dependent tumor rejection and that the **CD70** intracytoplasmic domain could mediate proliferative or surviving signals required for optimal effector/memory CD8 T cell generation.

LFA-3

L11 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1996:664627 CAPLUS
DOCUMENT NUMBER: 125:299413
TITLE: Compositions and methods for increasing the immunogenicity of tumor cells by administration of B7 and CD2-transfected cells
INVENTOR(S): Chen, Lieping; Hellstroem, Karl Erik
PATENT ASSIGNEE(S): Bristol-Myers Squibb Company, USA
SOURCE: Eur. Pat. Appl., 15 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 733373	A2	19960925	EP 1996-302009	19960322
EP 733373	A3	19980428		
R: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
CA 2172436	AA	19960924	CA 1996-2172436	19960322
JP 09012479	A2	19970114	JP 1996-105982	19960325

PRIORITY APPLN. INFO.: US 1995-409768 19950323

AB The present invention is directed to a method of inhibiting tumor cell growth. Tumor cells from a patient are transfected to express both B7 and a CD2 ligand on the surface of the transfected tumor cells and these cells are then readministered to the patient. The presence of the B7 and CD2 ligand mols. on the tumor cell surface stimulates a broad immunol. response against both the transfected and non-transfected tumor cells and results in the immunol. killing of localized and metastatic tumor cells. B7 and CD2 ligand transfected tumor cells, or cell membranes, serve as an immune enhancer to engender a potent immunol. response against the surface antigens present on the tumors cells.

L9 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:369788 CAPLUS
DOCUMENT NUMBER: 137:293462
TITLE: Transient blocking of both **B7**.1 (CD80) and **B7**.2 (CD86) in addition to **CD40** -CD40L interaction fully abrogates the immune response following systemic injection of adenovirus vector
AUTHOR(S): Ziller, C.; Stoeckel, F.; Boon, L.; Haegel-Kronenberger, H.
CORPORATE SOURCE: TRANSGENE, Strasbourg, 67082, Fr.
SOURCE: Gene Therapy (2002), 9(9), 537-546
CODEN: GETHEC; ISSN: 0969-7128
PUBLISHER: Nature Publishing Group
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Blockade of the CD40-CD40L and CD80/CD86-CD28 costimulatory pathways represents a strategy to inhibit the immune response against Ad (adenovirus) **vectors** designed for gene therapy applications. Since most previous studies have used a CTLA4-Ig fusion mol. binding to both CD80 and CD86, the resp. roles of these B7 mols. remained undefined. The authors have studied the effect of blocking monoclonal Abs (mAbs) directed against the **costimulatory mols**. CD40L, CD80 and CD86, alone or in different combinations, on the humoral and cellular immune responses against Ad. Groups of mice were transiently treated with each combination of blocking mAbs upon systemic injection of a first Ad **vector**. Combinations of anti-CD80 + anti-CD86 or anti-CD40L + anti-CD86 mAbs resulted in strong inhibition of the immune response against Ad. Using either of these mAb pairs, a second **vector** could be administered 1 mo after the first injection but with lower efficiency than in naive animals. Thus, CD86 stands as the pivotal B7 mol. involved in the development of the immune response against Ad. However, only the blockade of both CD80 and CD86 in addn. to CD40L fully inhibited the humoral and cellular responses against the Ad **vector** , such that readministration after 1 mo was as efficient as in naive animals. At the time of readministration, treated animals had regained their ability to mount a normal immune response to the second Ad **vector**, showing that tolerance was not induced.
REFERENCE COUNT: 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:167068 CAPLUS
DOCUMENT NUMBER: 134:324877
TITLE: The roles of MHC class II, **CD40**, and **B7** costimulation in CTL induction by plasmid DNA
AUTHOR(S): Chan, Kee; Lee, Delphine J.; Schubert, Amy; Tang, Chih Min; Crain, Brian; Schoenberger, Stephen P.; Corr, Maripat
CORPORATE SOURCE: Department of Medicine and The Sam and Rose Stein Institute for Research on Aging, University of California at San Diego, La Jolla, CA, 92093, USA
SOURCE: Journal of Immunology (2001), 166(5), 3061-3066
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists

DOCUMENT TYPE: Journal
LANGUAGE: English
AB DNA-based vaccines generate potent CTL responses. The mechanism of T cell stimulation has been attributed to plasmid-transfected dendritic cells. These cells have also been shown to express plasmid-encoded proteins and to become activated by surface marker up-regulation. However, the increased surface expression of **CD40** and **B7** on these dendritic cells is insufficient to overcome the need for MHC class II-restricted CD4+ T cell help in the priming of a CTL response. In this study, MHC class II-/- mice were unable to generate a CTL response following DNA immunization. This deficit in CTL stimulation by MHC class II-deficient mice was only modestly restored with CD40-activating Ab, suggesting that there were other elements provided by MHC class II-restricted T cell help for CTL induction. CTL activity was also augmented by coinjection with a **vector** encoding the costimulatory ligand B7.1, but not B7.2. These data indicate that dendritic cells in plasmid DNA-injected mice require conditioning signals from MHC class II-restricted T cells that are both CD40 dependent and independent and that there are different roles for **costimulatory mols**. that may be involved in inducing optimal CTL activity.
REFERENCE COUNT: 44 THERE ARE 44 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

=> DIS L7 1- IBIB ABS
YOU HAVE REQUESTED DATA FROM 8 ANSWERS - CONTINUE? Y/(N):Y
THE ESTIMATED COST FOR THIS REQUEST IS 19.32 U.S. DOLLARS
DO YOU WANT TO CONTINUE WITH THIS REQUEST? (Y)/N:Y

L7 ANSWER 1 OF 8 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:278014 CAPLUS
TITLE: Antitumor Immunity After Vaccination With B Lymphoma Cells Overexpressing a Triad of Costimulatory Molecules
AUTHOR(S): Briones, Javier; Timmerman, John M.; Panicalli, Dennis L.; Levy, Ronald
CORPORATE SOURCE: CA; D. L. Panicalli, Stanford, Division of Oncology, R. Levy, J. M. Timmerman, J. Briones, Stanford University School of Medicine, Therion Biologics Corporation, Cambridge, MA, USA
SOURCE: Journal of the National Cancer Institute (2003), 95(7), 548-555
CODEN: JNCIEQ; ISSN: 0027-8874
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Background: The **costimulatory mols**. **B7**-1, intercellular adhesion mol.-1 (**ICAM**-1), and leukocyte function-assocd. antigen-3 (**LFA**-3) play pivotal roles in the activation of T cells. We investigated whether in vivo vaccination with lymphoma cells infected with a recombinant, nonreplicating fowlpox (FP) virus encoding this triad of **costimulatory mols**. (TRICOM) could stimulate lymphoma-specific immunity. Methods: TRICOM-infected A20 B lymphoma cells were analyzed for expression of **B7**-1, **ICAM**-1, and **LFA**-3. Mice (10 per group) were vaccinated with irradiated A20 cells infected with either the TRICOM

vector or the wild-type FP virus (WT-FP), challenged with live A20 tumor cells, and followed for survival. Mice with established A20 tumors were also treated with irradiated TRICOM-infected A20 cells. Survival curves were compared with the log-rank statistic. The mechanism of the antitumor effect was studied by in vivo depletion of CD4+ and CD8+ T cells and in vitro cytotoxicity assays. All statistical tests were two-sided. Results: A20 tumor cells infected with TRICOM expressed high levels of **B7**-1, **ICAM**-1, and **LFA**-3. Mice vaccinated with irradiated TRICOM-infected A20 cells had prolonged survival relative to mice vaccinated with WT-FP-infected cells (80% vs. 20% survival at 110 days; P<.001). In mice with established tumors, tumor growth was slower in those treated with TRICOM-infected tumor cells than in those treated with WT-FP-infected cells, and this treatment provided a survival advantage (P<.001). Depletion of CD4+ or CD8+ T cells reduced the antitumor immunity provided by the tumor cell-TRICOM vaccine, and lymphocytes from vaccinated mice displayed in vitro cytotoxic activity toward A20 cells. Conclusions: Increasing expression of **costimulatory mols**. on B lymphoma cells by infection with a recombinant FP virus encoding **B7**-1, **ICAM**-1, and **LFA**-3 stimulates antitumor immune responses in vivo and may provide a novel strategy for treating patients with B-cell malignancies.

L7 ANSWER 2 OF 8 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:129253 CAPLUS
DOCUMENT NUMBER: 138:253285
TITLE: Selective Induction of High Avidity CTL by Altering the Balance of Signals from APC
AUTHOR(S): Oh, SangKon; Hodge, James W.; Ahlers, Jeffrey D.; Burke, Donald S.; Schlom, Jeffrey; Berzofsky, Jay A.
CORPORATE SOURCE: National Cancer Institute, and Laboratory of Tumor Immunology and Biology, Metabolism Branch, Molecular Immunogenetics and Vaccine Research Section, National Institutes of Health, Bethesda, MD, 20892, USA
SOURCE: Journal of Immunology (2003), 170(5), 2523-2530
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English

AB High avidity CTL are most effective at clearing viruses and cancer cells. Therefore, understanding the mechanisms involved in induction of high avidity CTL is crit. for effective vaccines. However, no vaccine approach to selectively induce high avidity CTL in vivo has been discovered. In a new approach, signals from MHC class I (signal 1) and costimulatory mols. (signal 2) were adjusted by varying Ag dose and by use of recombinant poxvirus expressing a triad of costimulatory mols. (**B7**-1, **ICAM**-1, and **LFA**-3), resp. Independent of CTL avidity, a strong signal 1 resulted in an increased frequency of CD8+ CTL. However, a strong signal 2 was necessary for the induction of high avidity CD8+ CTL that killed target cells more efficiently, and signal 2 played a more crucial role in the absence of a strong signal 1. Only CTL induced with strong signal 2 killed tumor cells endogenously expressing low levels of Ag. Signal 2 contributed to the induction of high avidity CD8+ CTL in both primary and secondary responses. Thus, although signal 2 has been known to increase the quantity of CTL response, in this study the authors show that it also improves the quality of CTL response. The data also suggested that dendritic cells play an important role in induction of high

avidity CD8+ CTL in vivo. This strategy to selectively induce higher avidity CTL may lead to more effective vaccines for viruses and cancer.

REFERENCE COUNT: 40 THERE ARE 40 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 3 OF 8 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:900432 CAPLUS
DOCUMENT NUMBER: 137:77486
TITLE: Enhanced activation of rhesus T cells by **vectors** encoding a triad of **costimulatory molecules** (**B7**-1, **ICAM**-1, **LFA**-3)
AUTHOR(S): Shankar, Pragyna; Schlom, Jeffrey; Hodge, James W.
CORPORATE SOURCE: Research Scholar's Program, NIH, Howard Hughes Medical Institute, Bethesda, MD, 20892, USA
SOURCE: Vaccine (2001), 20(5-6), 744-755
CODEN: VACCDE; ISSN: 0264-410X
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Since the rhesus is often used as a "gatekeeper" model for the evaluation of malaria and simian immunodeficiency virus (SIV)/HIV vaccines, the identification of strategies to enhance the activation of rhesus T cells would potentially aid in the generation of more potent vaccines directed against these infectious agents. Several mols. normally found on the surface of professional human APCs are capable of providing the second signals crit. for T cell activation: **B7**-1 (CD80), **ICAM**-1 (CD54), and **LFA**-3 (CD58). With the exception of B7, T cell **costimulatory mols**. in the rhesus have not been identified. We have recently designed and characterized both recombinant vaccinia and recombinant avipox **vectors** contg. the transgenes for a triad of human T cell **costimulatory mols**. (**B7**-1, **ICAM**-1, **LFA**-3; designated TRICOM). Here, we demonstrate the enhanced activation of rhesus T cells stimulated with rhesus APCs infected with TRICOM **vectors** in the presence of signal 1. Infection with TRICOM **vectors** led to significant improvement of APC capabilities in terms of redn. of the amt. of signal 1 needed to activate naive T cells, and redn. in the amt. of APCs required to activate T cells using a const. amt. of signal 1. Antibody blocking studies demonstrated that each of the three **costimulatory mol**. transgenes contributed to the enhanced proliferation of T cells. TRICOM-enhanced T cell activation was shown to correspond to increases in type 1 cytokines and a reduced level of apoptosis. TRICOM-infected autologous B cells from rhesus immunized with either an SIV vaccine or a malaria vaccine stimulated significantly greater levels of IFN-.gamma. in response to specific peptide than stimulation with uninfected autologous B cells or B cells infected with wild-type **vector**. The ability to augment immune responses using poxvirus-based vaccines contg. multiple **costimulatory mol**. transgenes can now be addressed in the rhesus macaque model.

REFERENCE COUNT: 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 4 OF 8 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:683489 CAPLUS

DOCUMENT NUMBER: 136:245917
TITLE: Technology evaluation: CEA-TRICOM, Therion Biologics Corp
AUTHOR(S): Morse, Michael A.
CORPORATE SOURCE: Department of Medicine, Duke University Medical Center, Durham, NC, 27710, USA
SOURCE: Current Opinion in Molecular Therapeutics (2001), 3(4), 407-412
CODEN: CUOTFO; ISSN: 1464-8431
PUBLISHER: PharmaPress Ltd.
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English
AB A review. Therion Biologics, the NCI and Aventis Pasteur are investigating CEA-TRICOM, a recombinant, pox virus-based vaccine that incorporates a triple dose of **costimulatory mols**. as well as the carcinoembryonic antigen (CEA) tumor antigen, for the potential treatment of colorectal cancer. CEA-TRICOM is designed to stimulate and strengthen the body's immune system to kill colorectal cancer cells. CEA-TRICOM is administered in a priming and boosting protocol using two unique pox virus **vectors**, rV-CEA-TRICOM (recombinant vaccinia **vector**) and rF-CEA-TRICOM (recombinant fowlpox **vector**). The TRICOM component of both rV-CEA-TRICOM and rF-CEA-TRICOM comprises three **costimulatory mol**. transgenes (**B7**-1, **ICAM**-1 and **LFA**-3) known to elicit strong cellular immune responses necessary for complete tumor destruction. In preclin. studies conducted by the NCI and Therion, researchers have demonstrated that this combination of three **costimulatory mols**. dramatically boosts the immune response to eradicate cancer in murine models. In Feb. 2001, Therion Biologics and the NCI initiated a phase I trial of CEA-TRICOM. The phase I trial of CEA-TRICOM is designed to demonstrate proof-of-principle for using multiple **costimulatory mols**. in conjunction with a tumor antigen to improve the strength of cellular immune responses. It is a multistage, dose-escalation study that will assess the safety and immunol. effects of CEA-TRICOM in up to 42 patients who have advanced metastatic colorectal cancer. Subjects will receive rF-CEA-TRICOM alone, rV-CEA-TRICOM followed by booster vaccinations with rF-CEA-TRICOM or rV-CEA-TRICOM followed by rF-CEA-TRICOM and GM-CSF adjuvant. The primary measure of immune response will be the level of CEA-specific T-cells stimulated by vaccination, with levels of CEA-expressing tumor cells in the blood used as a potential secondary measure of treatment effect.
REFERENCE COUNT: 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE
FORMAT

L7 ANSWER 5 OF 8 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:431014 CAPLUS
DOCUMENT NUMBER: 135:179366
TITLE: Synergy of vaccine strategies to amplify antigen-specific immune responses and antitumor effects
AUTHOR(S): Grosenbach, Douglas W.; Barrientos, Jacqueline C.; Schlom, Jeffrey; Hodge, James W.
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892-1750, USA
SOURCE: Cancer Research (2001), 61(11), 4497-4505
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Several different vaccine strategies have been evaluated and combined to amplify T-cell responses toward induction of antitumor immunity. The model tumor antigen used was carcinoembryonic antigen (CEA). While initial T-cell activation studies were conducted in conventional mice, combined vaccine strategy studies and antitumor studies were conducted in transgenic mice in which CEA is expressed in normal gastrointestinal tissue and CEA protein is found in sera. The studies reported here demonstrate: (a) A recombinant avipox (fowlpox, rF) **vector** expressing the signal 1 (CEA) and the **B7**-1 **costimulatory mol**. transgenes (designated rF-CEA/**B7**-1) is more potent in inducing CEA-specific T-cell responses than rF-CEA; one administration of recombinant fowlpox **vector** expressing CEA and three different **costimulatory mol**. transgenes (**B7**-1, **ICAM**-1, **LFA**-3, designated rF-CEA/TRICOM) was more potent in inducing CEA-specific T-cell responses than four vaccinations with rF-CEA or two vaccinations with rF-CEA/**B7**-1. Moreover, up to four vaccinations with rF-CEA/TRICOM induced greater CEA-specific T-cell responses with each vaccination. (b) A diversified prime and boost strategy using a prime with a recombinant vaccinia **vector** expressing CEA and the triad of **costimulatory mols**. (designated rV-CEA/TRICOM) and a boost with rF-CEA/TRICOM was more potent in inducing CEA-specific T-cell responses than the repeated use of rF-CEA/TRICOM alone. (c) The addn. of granulocyte macrophage colony-stimulating factor (GM-CSF) to the rF-CEA or rF-CEA/TRICOM vaccinations via the simultaneous administration of a rF-GM-CSF **vector** enhanced CEA-specific T-cell responses. These strategies (TRICOM/diversified prime and boost/GM-CSF) were combined to treat CEA-expressing carcinoma liver metastases in CEA-transgenic mice; vaccination was initiated 14 days post-tumor transplant. Antitumor effects in terms of survival and CD8+ and CD4+ responses specific for CEA were also obsd. in this CEA-transgenic mouse model. These studies demonstrate that the use of cytokines and diversified prime and boost regimens can be combined with the use of recombinant **vectors** expressing signal 1 and multiple **costimulatory mols**. to further amplify T-cell responses toward more effective vaccine strategies.

L7 ANSWER 6 OF 8 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:354386 CAPLUS
DOCUMENT NUMBER: 135:120924
TITLE: Enhanced activation of human T cells via avipox **vector**-mediated hyperexpression of a triad of **costimulatory molecules** in human dendritic cells
AUTHOR(S): Zhu, MingZhu; Terasawa, Hiroshi; Gulley, James; Panicali, Dennis; Arlen, Philip; Schlom, Jeffrey; Tsang, Kwong Y.
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892-1750, USA
SOURCE: Cancer Research (2001), 61(9), 3725-3734
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB T-cell activation usually requires at least 2 signals. The first signal is antigen-specific, and the second signal(s) involves the interaction of a T-cell **costimulatory mol**.(s) on the antigen-presenting cell (APC) with its ligand on the T cell. Dendritic cells (DCs) are the most potent APCs, attributable, in part, to their expression of several T-cell **costimulatory mols**.
Human DCs generated in vitro, however, will vary in methods of generation and maturation and in terms of expression of different phenotypic markers-including **costimulatory mols**.-among different donors. The authors report here that a recombinant avipox (fowlpox, rF) **vector** has been constructed that can efficiently express the transgenes for 3 human T-cell **costimulatory mols**. (**B7**-1, **ICAM**-1, and **LFA**-3) as a result of individual early avipox promoters driving the expression of each transgene. This triad of **costimulatory mols**. (designated TRICOM) was selected because each has an individual ligand on T cells and each has been shown previously to prime a unique signaling pathway in T cells. The authors report here that rF-TRICOM can efficiently infect human DCs of different states of maturity and hyperexpress each of the 3 **costimulatory mols**. on the DC surface without affecting other DC phenotypic markers. Infection of influenza or human papilloma virus 9-mer peptide-pulsed DCs from different individuals, or at different stages of maturity with rF-TRICOM, resulted in enhanced activation of T cells from peripheral blood mononuclear cells of autologous donors after 24 h of incubation with DCs. This enhanced activation was analyzed by both titrating the peptide and differing the DC:effector cell ratios. No effect was obsd. using the control wild-type avipox **vector**. No increase in apoptosis was obsd. in T cells hyperstimulated with the TRICOM **vector**, and no decrease in interleukin-12 prodn. was seen in lipopolysaccharide-stimulated DCs infected with rF-TRICOM. Antibody-blocking expts. demonstrated that enhanced T-cell activation by TRICOM was attributed to each of the 3 **costimulatory mols**. Peptide-pulsed, rF-TRICOM-infected DCs were also shown to be more effective than peptide-pulsed DCs in activating T cells to 9-mer peptides derived from 2 relatively weak "self" immunogens, i.e., human prostate-specific antigen and human carcinoembryonic antigen. These studies thus demonstrate for the first time that a **vector** that can simultaneously hyperexpress 3 **costimulatory mols**. can be used to efficiently infect human DCs, leading to enhanced peptide-specific T-cell activation. The use of this approach for in vitro studies and clin. applications in

immunotherapy is discussed.
REFERENCE COUNT: 62 THERE ARE 62 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE
FORMAT

TITLE: Enhancing the potency of peptide-pulsed antigen presenting cells by **vector**-driven hyperexpression of a triad of **costimulatory molecules**

AUTHOR(S): Hodge, J. W.; Grosenbach, D. W.; Rad, A. N.; Giuliano, M.; Sabzevari, H.; Schlom, J.

CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, National Institutes of Health, Bethesda, MD, 20892-1750, USA

SOURCE: Vaccine (2001), 19(25-26), 3552-3567
CODEN: VACCDE; ISSN: 0264-410X

PUBLISHER: Elsevier Science Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Recombinant orthopox **vectors** [both replication-defective fowlpox (rF), and replication competent vaccinia (rV)] have been developed that simultaneously express 3 T-cell **costimulatory mol.** transgenes. The constituents of this triad of **costimulatory mols.** (designated TRICOM) are **B7**-1, **ICAM**-1, and **LFA**-3. The authors have previously shown that infection of murine dendritic cells (DCs) with TRICOM **vectors** increases their level of expression of the triad of **costimulatory mols** . and enhances the efficacy of DCs to activate T cells. While DCs are arguably the most potent antigen presenting cell (APC), limitations clearly exist in their use due to the level of effort and cost for their generation. The studies reported here demonstrate that a generic APC population, murine splenocytes, can be made markedly more efficient as APCs by infection with either rF-TRICOM or rV-TRICOM **vectors**. Infection of splenocytes with either TRICOM **vector** led to improvement of APC capabilities in terms of: (1) enhancement of mixed lymphocyte reactions; (2) a redn. in the amt. of signal 1 to activate naive T cells; and (3) a redn. in the amt. of APCs required to activate T cells using a const. amt. of signal 1. TRICOM-enhanced T-cell activation was shown to correspond to increases in type-1 cytokines and a reduced level of apoptosis, compared with T cells activated with uninfected or control **vector**-infected splenocytes. In vitro and in vivo expts. compared DCs with TRICOM-infected splenocytes. Infection of splenocytes with TRICOM **vectors** markedly enhanced their ability to activate T cells to levels approaching that of DCs. These studies thus demonstrate for the first time that an abundant and accessible population of APCs obtainable without lengthy culture or the use of costly exogenous cytokines (in contrast to that of DCs) can be made more potent as APCs with the use of **vectors** that express a triad of **costimulatory mols**.

REFERENCE COUNT: 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 8 OF 8 CAPLUS

L7 ANSWER 8 OF 8 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1999:778296 CAPLUS
DOCUMENT NUMBER: 132:77584
TITLE: A triad of costimulatory molecules synergize to amplify T-cell activation
AUTHOR(S): Hodge, James W.; Sabzevari, Helen; Yafal, Alicia Gomez; Gritz, Linda; Lorenz, Matthias G. O.; Schlom, Jeffrey
CORPORATE SOURCE: Laboratory of Tumor Immunology and Biology, National Cancer Institute, NIH, Bethesda, MD, 20892, USA
SOURCE: Cancer Research (1999), 59(22), 5800-5807
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: AACR Subscription Office
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The activation of a T cell has been shown to require 2 signals via mols. present on professional antigen-presenting cells: signal 1, via a peptide/MHC complex; and signal 2, via a **costimulatory mol**. Here, the role of 3 **costimulatory mols**. in the activation of T cells was examd. Poxvirus (vaccinia and avipox) **vectors** were used because of their ability to efficiently express multiple genes. Murine cells provided with signal 1 and infected with either recombinant vaccinia or avipox **vectors** contg. a TRIad of **COstimulatory Mols**. (**B7**-1/**ICAM**-1/ **LFA**-3, designated TRICOM) induced the activation of T cells to a far greater extent than cells infected with any 1 or 2 **costimulatory mols**. Despite this T-cell "hyperstimulation" using TRICOM **vectors**, no evidence of apoptosis above that seen using the B7-1 **vector** was obsd. Results using the TRICOM **vectors** were most dramatic under conditions of either low levels of first signal or low stimulator cell:T-cell ratios. Expts. using a 4-gene construct also showed that TRICOM recombinants can enhance antigen-specific T-cell responses in vivo.
These studies thus demonstrate for the first time the ability of **vectors** to introduce 3 **costimulatory mols**. into cells, thereby activating both CD4+ and CD8+ T-cell populations to levels greater than those achieved with the use of only 1 or 2 **costimulatory mols**. This new threshold of T-cell activation has broad implications in vaccine design and development.
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE
FORMAT

L5 ANSWER 1 OF 9 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:278014 CAPLUS
TITLE: Antitumor Immunity After Vaccination With B Lymphoma Cells Overexpressing a Triad of Costimulatory Molecules
AUTHOR(S): Briones, Javier; Timmerman, John M.; Panicalli, Dennis L.; Levy, Ronald
CORPORATE SOURCE: CA; D. L. Panicalli, Stanford, Division of Oncology, R. Levy, J. M. Timmerman, J. Briones, Stanford University School of Medicine, Therion Biologics Corporation, Cambridge, MA, USA
SOURCE: Journal of the National Cancer Institute (2003), 95(7), 548-555
CODEN: JNCIEQ; ISSN: 0027-8874
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Background: The **costimulatory mols.** **B7**-1, intercellular adhesion mol.-1 (ICAM-1), and leukocyte function-assocd. antigen-3 (**LFA**-3) play pivotal roles in the activation of T cells. We investigated whether in vivo vaccination with lymphoma cells infected with a recombinant, nonreplicating fowlpox (FP) virus encoding this triad of **costimulatory mols.** (TRICOM) could stimulate lymphoma-specific immunity. Methods: TRICOM-infected A20 B lymphoma cells were analyzed for expression of **B7**-1, ICAM-1, and **LFA**-3. Mice (10 per group) were vaccinated with irradiated A20 cells infected with either the TRICOM **vector** or the wild-type FP virus (WT-FP), challenged with live A20 tumor cells, and followed for survival. Mice with established A20 tumors were also treated with irradiated TRICOM-infected A20 cells. Survival curves were compared with the log-rank statistic. The mechanism of the antitumor effect was studied by in vivo depletion of CD4+ and CD8+ T cells and in vitro cytotoxicity assays. All statistical tests were two-sided. Results: A20 tumor cells infected with TRICOM expressed high levels of **B7**-1, ICAM-1, and **LFA**-3. Mice vaccinated with irradiated TRICOM-infected A20 cells had prolonged survival relative to mice vaccinated with WT-FP-infected cells (80% vs. 20% survival at 110 days; P<.001). In mice with established tumors, tumor growth was slower in those treated with TRICOM-infected tumor cells than in those treated with WT-FP-infected cells, and this treatment provided a survival advantage (P<.001). Depletion of CD4+ or CD8+ T cells reduced the antitumor immunity provided by the tumor cell-TRICOM vaccine, and lymphocytes from vaccinated mice displayed in vitro cytotoxic activity toward A20 cells. Conclusions: Increasing expression of **costimulatory mols.** on B lymphoma cells by infection with a recombinant FP virus encoding **B7**-1, ICAM-1, and **LFA**-3 stimulates antitumor immune responses in vivo and may provide a novel strategy for treating patients with B-cell malignancies.